FEBS Letters 584 (2010) 1509–1514

FEBS Letters
journal homepage: www.FEBSLetters.org

The structure of Get4 reveals an α-solenoid fold adapted for multiple interactions in tail-anchored protein biogenesis

Gunes Bozkurt [a], Klemens Wild [a], Stefan Amlacher [a], Ed Hurt [a], Bernhard Dobberstein [b], Irmgard Sinning [a,*]

[a] Heidelberg University Biochemistry Center (BZH), Im Neuenheimer Feld 328, D-69120 Heidelberg, Germany
[b] Zentrum für Molekulare Biologie der Universität Heidelberg (ZMBH), DKFZ – ZMBH Allianz, Im Neuenheimer Feld 282, D-69120 Heidelberg, Germany

ARTICLE INFO

Article history:
Received 19 February 2010
Revised 24 February 2010
Accepted 25 February 2010
Available online 3 March 2010

Edited by Richard Cogdell

Keywords:
Get pathway
Posttranslational targeting
Tail-anchored membrane protein insertion
TPR-like fold

ABSTRACT

Tail-anchored proteins play important roles in protein translocation, membrane fusion and apoptosis. They are targeted to the endoplasmic reticulum membrane via the guided-entry of tail-anchored proteins (Get) pathway. We present the 2 Å crystal structure of Get4 which participates in early steps of the Get pathway. The structure shows an α-solenoid fold with particular deviations from the regular pairwise arrangement of α-helices. A conserved hydrophobic groove accommodates the flexible C-terminal region *in trans*. The structural organization of the Get4 helical hairpin motifs provides a scaffold for protein–protein interactions in the Get pathway.

1. Introduction

Tail-anchored (TA) membrane proteins represent a significant percentage of the eukaryotic membrane proteome [1]. They play important roles in various cellular processes, including membrane fusion (e.g. SNARE proteins), protein translocation (e.g. Sec61ß), and apoptosis (e.g. Bcl2) [2–4]. In contrast to N-terminal signal sequences employed by typical SRP (signal recognition particle) substrates [5,6], TA proteins carry their targeting signal within a single transmembrane domain (TMD) at the C-terminus [7]. Therefore, the targeting signal is exposed to the cytosol after the release from the ribosome and TA proteins are post-translationally targeted. Recently, Get3 was identified as a cytosolic ATPase that binds and targets cargo proteins (TA proteins) in a nucleotide dependent manner to the ER membrane [8,9]. Several structures of Get3 in different nucleotide loads shed light on the catalytic cycle of the Get3 ATPase as well as on the TA protein binding site [10–14]. Genetic and biochemical studies identified four additional components in the guided-entry of tail-anchored proteins (Get) pathway: Get1, Get2, Get4, and Get5. Get1/Get2 provide the receptor at the ER membrane required for TA protein targeting [15] and interact with Get3 [16]. Get4 (YOR164C in yeast) and Get5 (Mdy2p in yeast) interact with Get3 in the cytosol and in yeast form the TMD recognition complex [16]. Get4 and Get5 were found associated with ribosomes [17]. Therefore, both proteins are suggested to play a role in the cargo loading step. Knock-out yeast strains of get4 and get5 (*Δget4* and *Δget5*) show severe defects in TA protein biogenesis such as a localization defect of the cargo protein Sed5 [16]. Although the Get pathway has recently received quite some attention, our understanding of the individual steps and protein interactions along this pathway is still rather limited.

In order to add to the molecular details of TA protein targeting, we determined the crystal structure of Get4 from a thermophilic fungus at 2 Å resolution. The structure provides insights in the adaptation of the α-2-solenoid fold to allow interactions to Get4 with components of the Get pathway.

2. Materials and methods

2.1. Cloning, expression and purification

The gDNA of the thermophilic fungus *Chaetomium thermophilum* (*C. therm.*) was sequenced (Amlacher and Hurt, in preparation). The gene encoding residues 1–329 of *C. therm.* Get4 was amplified from a cDNA preparation and cloned into the pET-24d vector using the NcoI/BamHI sites. All DNA constructs were sequenced by AGOWA, Berlin. Native and selenomethionine (SeMet) labelled Get4 were over-expressed in *Escherichia coli* strain BL21 (DE3) Rosetta (Novagen) at 37 °C, the latter one in a supplemented M9 medium with 1 mM IPTG induction at an OD_{600} 0.6. Cell pellets were resuspended in a buffer consisting of 10 mM Tris–Cl pH 8.0, 500 mM

* Corresponding author. Fax: +49 6221 544790.
E-mail address: irmi.sinning@bzh.uni-heidelberg.de (I. Sinning).

0014-5793/$36.00 © 2010 Federation of European Biochemical Societies. Published by Elsevier B.V. All rights reserved.
doi:10.1016/j.febslet.2010.02.070

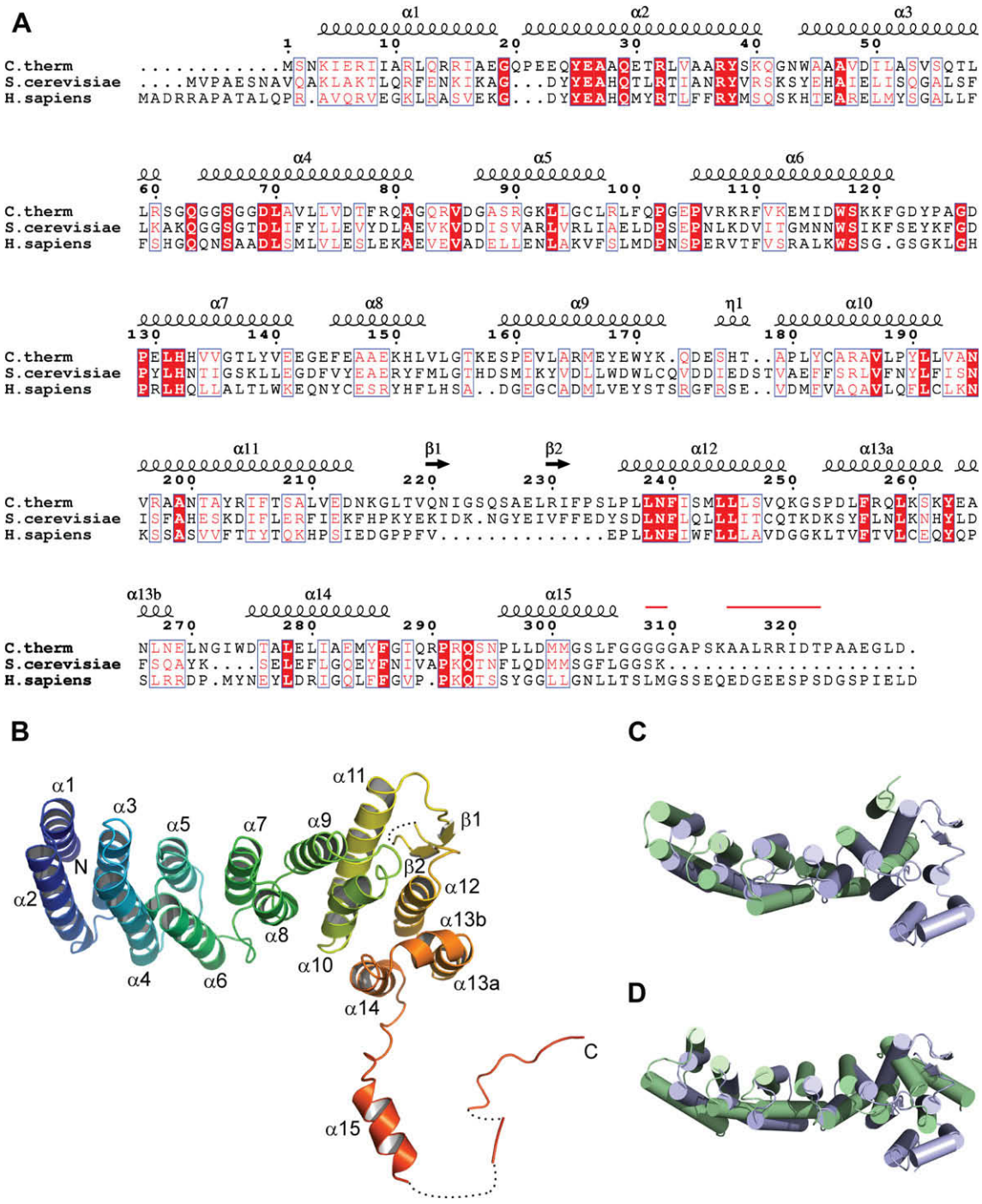

Fig. 1. Structure of Get4. (A) Multiple sequence alignment of Get4 homologs from *C. therm.*, *S. cerevisiae*, and *H. sapiens*. Secondary structure elements of *C. therm.* Get4 are indicated above the sequence. Numbering is according to the sequence of *C. therm*. Get4. Red lines indicate the residues interacting *in trans* with a symmetry related molecule (see Fig. 3A). (B) Ribbon representation of the *C. therm.* Get4 structure. Coloring is done in a ramp from blue (N-terminus) to red (C-terminus). N- and C-termini, and secondary structure elements are labelled. Dotted lines indicate disordered regions in the crystal structure. *C. therm.* Get4 (in light blue) is superimposed with structural homologs that share the α-2-solenoid fold in (C) the vacuolar protein sorting-associated protein 35, Vsp35 (PDB entry code 2r17-chainD residues 522–780; green) and (D) the adaptor protein complex AP-2 α-subunit (PDB entry code 1gw5-chainA residues 253–508; green).

NaCl, 20 mM imidazol, 1 mM $MgCl_2$, 1 mM DTT). Cells were lysed using a Microfluidizer M110L (Microfluidics). The protein was purified using a His-Trap HP column (GE Healthcare) followed by size exclusion chromatography (Superdex 75 26/60, GE Healthcare) in a buffer consisting of 10 mM Tris–Cl pH 8.0, 150 mM NaCl, 1 mM $MgCl_2$ and 1 mM DTT. The same purification protocol was used for the SeMet-labelled protein.

2.2. Crystallization and structure determination

Native and SeMet crystals of *C. therm.* Get4 were grown at 4 °C. The crystallization conditions, structure determination using the single wavelength anomalous dispersion (SAD) method as well as refinement are described in the Supplementary data. Atomic coordinates and structure factors for the crystal structure of *C. therm.*

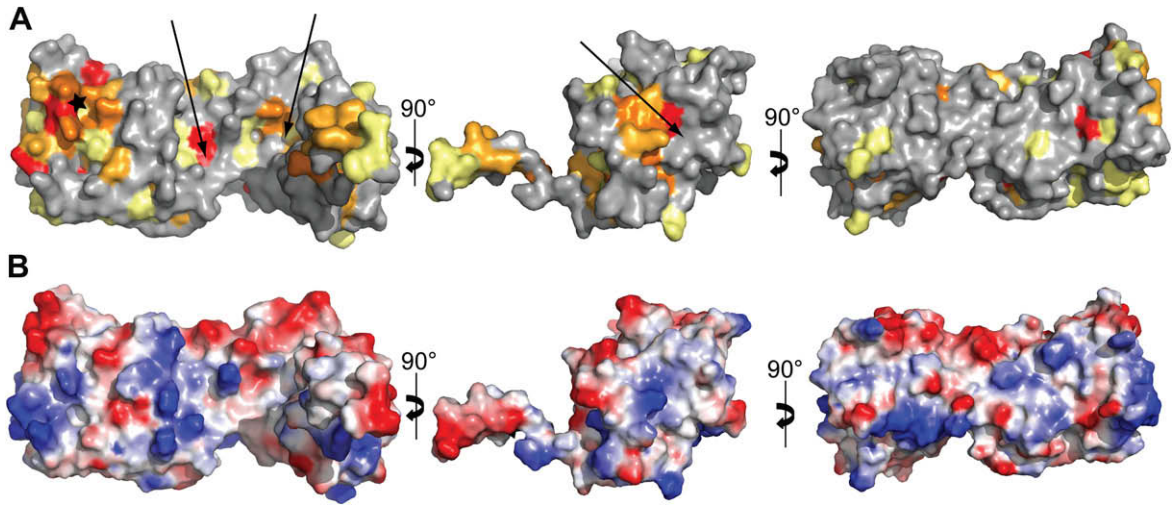

Fig. 2. Surface analysis of Get4. (A) The degree of sequence conservation within Get4 proteins is mapped onto the surface of the structure in three orientations: the convex surface (left panel), the tip (middle) and the concave surface (right). Red, dark orange, orange and yellow indicate residues that are highly or partially conserved, respectively. Clefts are indicated by arrows, the conserved patch at helices α2 and α4 is indicated by a star. (B) Electrostatic surface potential is shown for the same orientations as in (A). The molecular surface is colored blue and red according to positive and negative electrostatic potential, respectively.

Get4 have been deposited in the Protein Data Bank with the accession code 3LPZ.

3. Results and discussion

3.1. Get4 has an α-2-solenoid fold

In order to understand the role of Get4 in the Get pathway in more detail, we expressed, purified and crystallized full length Get4 from the thermophilic fungus *C. therm.* Get4 shares 29% identity and 50% homology with *Saccharomyces cerevisiae* Get4 (24% and 45% with *Homo sapiens* Get4, respectively) (Fig. 1A). Get4 is a stable monomer in solution (not shown). The structure was determined at 2 Å resolution using SAD. Crystallographic statistics are given in Supplementary Table 1. Get4 has an elongated, slightly curved shape with dimensions of $70 \times 30 \times 40$ Å (Fig. 1B). The structure is well ordered with the exception of a short flexible loop (residues 217 and 218) and a disordered region at the C-terminus (residues 306–314 and 324–329).

Get4 comprises 14 α-helices arranged pairwise with an α-2-solenoid fold [18], and an additional α-helix in the C-terminal region. The N-terminal region of Get4 (residues 1–156, helices α1–α8) shows a slight curvature and its architecture is similar to a TPR (Tetratrico Peptide Repeat)-like fold [19,20]. Vps35 [21] was identified as the closest structural homologue of Get4 by the program DALI [22] with a root mean square deviation (rmsd) of 2.7 Å over 126 residues (Fig. 1C). Other structural homologues include the α-subunit of the endocytic AP-2 complex [23] (rmsd of 3.3 Å over 107 residues; Fig. 1D), cullin-homolog1 [24] (rmsd 3.3 Å over 111 residues), Sec17 [25] (rmsd of 3.5 Å over 107 residues), and protein phosphatase 2A [26] (rmsd 3.4 Å over 109 residues). The regular arrangement of the first three helical hairpin repeats (α1–α2, α3–α4, α5–α6) in Get4 is distorted at the fourth repeat by an insertion in the loop connecting α6 and α7 (Fig. 1A and B). As a consequence, α7 and α8 do not pack as tightly to the preceding repeat as observed for the first three repeats. In addition, α8 is shorter than the other helices and small residues in the interface enable a tight packing of α7 and α8 which leads to particular deviations from a regular arrangement. The distortion continues in the C-terminal part of Get4 (residues 157–329) with six α-helices. Briefly, α9 is tilted and shifted away from α7, and α10 contains a proline kink. The insertion of a ß-hairpin between α11 and α12 prevents the addition of the following helix pair (α13–α14), which instead is attached on the side of the α-2-solenoid. Overall, the deviation from the pairwise helical arrangement introduced by the fourth repeat creates a pronounced cleft at the convex surface of Get4, while the insertion of a ß-hairpin and the addition of α13–α14 on the side lead to a packing defect which leaves the binding site of one helix (in a regular α-2-solenoid array) empty. The Get4 C-terminus (residues 291–329) is not part of the α-2-solenoid fold and appears to be flexible. The regions comprising helix α15 (residues 296–305) and residues 308–310 and 315–323 interact *in trans* with two different symmetry related molecules (see below; Supplementary Fig. 1).

3.2. Get4 surface analysis identifies conserved protein interaction sites

Get4 was shown to interact with Get5 [17,27] and the Get4/5 complex has been suggested to bind to ribosomes [17] as well as to Get3 [16]. In order to investigate the structure of Get4 for protein interaction sites, the conserved surface areas (Fig. 2A) and the electrostatic surface potential (Fig. 2B) were analysed. Get4 is slightly acidic with a pI of 6.2 and striking differences are observed between the concave and convex surfaces for both, conservation and electrostatic surface potential. The concave surface of Get4 (Fig. 2A, right panels) is not conserved. The convex surface (left panels) however displays conserved regions in the C-terminal part, as well as at the first two helical repeats at the N-terminus. This suggests that the convex surface of Get4 might be used for protein interactions, e.g. with Get3 and/or Get5. Close structural homologues of Get4 such as Vps35 and AP-2 both employ the concave surface for protein interactions [21,23]. Similarly, TPR proteins use their concave surface for protein–protein interactions [28–30], but also their convex side e.g. in the Fis1/Caf4 structure [31].

The conserved region in the N-terminal part is formed by α2 and α4, which includes also several charged residues (Figs. 1A and 2B). Interestingly, these residues are involved in a crystal contact which could indicate an interaction site (Supplementary Fig. 1). The N-terminal part of Get4 was shown to be involved in Get3 interaction in a yeast two-hybrid assay [32]. Our analysis of the Get4 structure supports these data and suggests that the proposed protein interaction site may localize at α2 and α4 on the

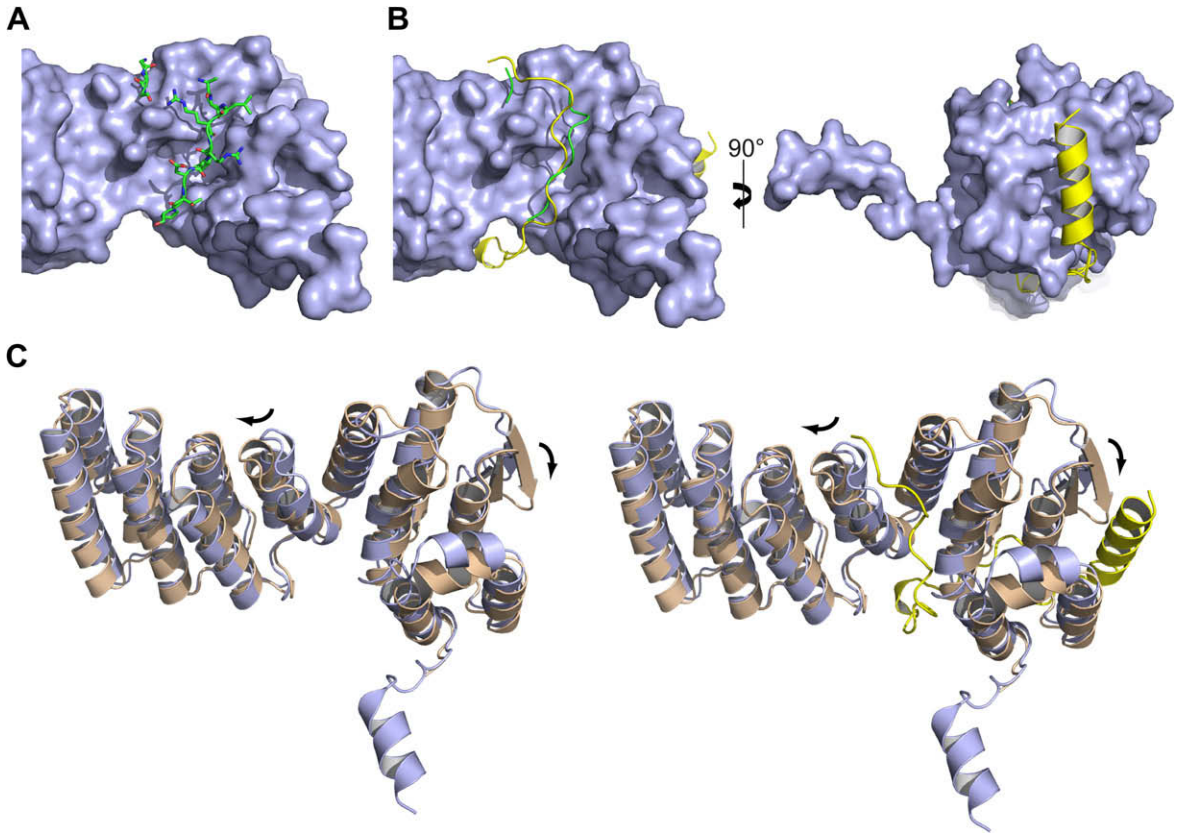

Fig. 3. Analysis of Get4 interaction sites (A). C-terminal region of *C. therm.* Get4 interacts *in trans* with part of the central cleft. The elongated interacting peptides comprising residues 308–310, and 315–323 are represented as sticks (green). Get4 is shown in a surface representation (light blue). (B) Superposition of *C. therm.* Get4 (surface in light blue) with the *S. cer.* Get5 from the Get4/Get5 complex (PDB entry code 2wpv; ribbon representation in yellow). The *in trans* interacting peptides (same as in A) are shown as a ribbon (green). (C) Superposition of *C. therm.* Get4 (blue) with the *S. cer.* Get4/Get5 complex (Get4 in wheat, Get5 in yellow, PDB entry code 2wpv). Conformational changes in Get4 upon Get5 binding are indicated by arrows (see text). The C-terminus of *C. therm.* Get4 is shown up to α15, which is not resolved in the *S. cer.* Get4 structure.

convex side of the helical array. In addition to this conserved patch a large cleft is present in the center of the convex surface which is also conserved (Fig. 2, left panels). Closer inspection reveals a bipartite structure with one part (to the right) being mainly hydrophobic while the left part is more polar with a number of positively charged residues. In addition, a second cleft localizes at the tip of Get4 (Fig. 2, middle panels). The insertion of a ß-hairpin after α11 seems to cause a packing defect which creates a conserved cleft formed by mainly hydrophobic residues. Taken together, the surface analysis suggests that the concave surface of Get4 might not be involved in protein interactions, while the convex surface and the cleft at the tip display properties typical for protein interaction sites. They might therefore participate in the interactions with Get3 and Get5.

3.3. The Get4 C-terminus occupies the Get5 binding site

During refinement of the Get4 crystal structure an elongated electron density was observed in the hydrophobic cleft at the convex surface. The polar part of the cleft is however empty. An elongated peptide could be fitted into the electron density representing part of the flexible C-terminal region of a symmetry related Get4 molecule (residues 308–310 and 315–323) (Fig. 3A; Supplementary Fig. 1). Due to the size and shape of the hydrophobic cleft we first hypothesized that it might be involved in TA protein binding. When we modeled the TMD of Sec61β [33] into the cleft we found that Get4 could indeed accommodate an α-helix (not shown). Get4 has been shown to interact with Get3 and Get5 [16,27] and therefore, the cleft might contribute to these interactions. Recently, the structure of *S. cerevisiae* Get4 in complex with the N-terminal region of Get5 was reported [32]. Here the central hydrophobic cleft in Get4 accommodates an elongated peptide from Get5 corresponding to the loop region after the short helix α2 (Fig. 3B, left panel). Superposition of both structures shows that the C-terminal region of Get4 in our structure (residues 308–310 and 315–323) superimposes well with the Get5 loop (residues 41–54) (Fig. 3B, left panel). Notably, the Get4 and Get5 peptides both bind to the hydrophobic cleft, but with opposite orientations and their sequences are not conserved. The more polar part of the cleft is empty in both cases.

The Get4 surface analysis (see above) identified a second conserved cleft on the tip (Fig. 2, middle panels). The C-terminal helix α15 binds in close vicinity to this cleft *in trans* (Supplementary Fig. 1). In our initial analysis we hypothesized that α15 could occupy this cleft as its surface characteristics match the properties of the cleft. However, in the *S. cer.* Get4/5 complex this cleft is occupied by Get5 α1 (Fig. 3B, right panel) [32]. This helix has amphipathic properties similar to Get4 α15 in our structure, and buries its hydrophobic side in the interface with Get4. This interaction with Get5 complements the helical hairpin arrangement of Get4 and "heals" the packing defect in the Get4 structure (Fig. 3B, right panel). We therefore speculate that in the absence of Get5, Get4 α15 might occupy this cleft, and is displaced by Get5 α1 upon formation of the heterodimer.

3.4. Get4/5 complex formation involves distinct adjustments

Comparison of isolated Get4 with the Get4/5 complex [32] shows that both Get4 structures are very similar, underlining the high conservation of the α-2-solenoid fold (rmsd of 1.53 Å over

260 residues between *C. therm.* Get4 and *S. cer.* Get4/Get5) (Fig. 3C). Only minor conformational changes are induced in Get4 upon binding of Get5, which localize to the C-terminal part of the structure (superposition of α1–α8: rmsd 1.27 Å over 149 residues, and of α9–α14: rmsd 1.37 Å over 113 residues). Therefore, Get4 can accommodate Get5 without large conformational changes. Two specific adjustments in the Get4/5 complex are however observed: the ß-hairpin in Get4 is stabilized, and moves by about 3 Å towards Get5 α1; the loop region between α7 and α8 moves by about 4 Å towards the N-terminal region which further opens the hydrophobic binding cleft for Get5. Taken together, the α-2-solenoid fold of Get4 provides a stable scaffold for the interaction with different proteins. The deviations from a regular helix arrangement in Get4 create two clefts that are used in Get5 binding.

In the Get4/5 structure [32] the C-terminal region of Get4 (residues 293–312) including α15, which is conserved in all Get4 homologues (Figs. 1A and 2) is not resolved. Therefore α15, which is connected to the α-2-solenoid scaffold by a flexible linker in the Get4 structure (this work), is detached in the Get4/5 complex and available to interact with another component of the Get pathway. The α-helical subdomain of Get3 which is thought to form the TA protein binding site comprises a number of amphipathic helices similar to α15. Whether α15 may participate in TA protein binding by Get3 remains to be seen.

4. Conclusions

The Get pathway is essential for efficient targeting of TA proteins to the ER [15]. Get4 and Get5 participate in the biogenesis of TA proteins and function upstream of Get3 [16]. Get4 has an α-2-solenoid fold and a flexible C-terminal region with a conserved amphipathic helix. The structure of Get4 provides a robust scaffold with conserved interaction sites that allow to assemble larger complexes in the early steps of the Get pathway, e.g. Get3, Get5 and/or Sgt2 [16,27,34]. Get4 forms a stable complex with Get5 in which the N-terminal helix of Get5 binds to the tip of Get4 and complements the α-2-solenoid fold. An elongated peptide of Get5 occupies a hydrophobic binding cleft [32], which can also accommodate other peptides even in the opposite orientation (this work). The presence of an extended water network in the vicinity of the interacting peptides in both cases raises questions on the specificity of the observed interactions. Since a large part of the cleft is empty also in the Get4/5 complex, it could be employed in another interaction of Get4/5 e.g. with Get3 or a Get3/TA protein complex. More biochemical experiments are needed to dissect the interaction of the components of the Get pathway.

Acknowledgements

We thank Jürgen Kopp and Claudia Siegmann of the BZH-CellNetworks crystallization platform, Basak Arslan and Fabio Villardi (ZMBH) for experimental support, Sebastian Falk for ITC experiments and Gert Bange for technical advice and discussions. We gratefully acknowledge access to the beamlines ID14-4 and ID23-1 at the ESRF (Grenoble, France) and thank the beamline scientists for excellent support. This work was supported by a Fellowship of the Boehringer Ingelheim Fonds, the Graduierten-Kolleg GRK1188, a collaborative research grant of the Deutsche Forschungsgesellschaft (SFB638) and the Cluster of Excellence:CellNetworks.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.febslet.2010.02.070.

References

[1] Kalbfleisch, T., Cambon, A. and Wattenberg, B.W. (2007) A bioinformatics approach to identifying tail-anchored proteins in the human genome. Traffic 8, 1687–1694.
[2] Borgese, N., Brambillasca, S., Soffientini, P., Yabal, M. and Makarow, M. (2003) Biogenesis of tail-anchored proteins. Biochem. Soc. Trans. 31, 1238–1242.
[3] Borgese, N., Brambillasca, S. and Colombo, S. (2007) How tails guide tail-anchored proteins to their destinations. Curr. Opin. Cell Biol. 19, 368–375.
[4] Rabu, C., Schmid, V., Schwappach, B. and High, S. (2009) Biogenesis of tail-anchored proteins: the beginning for the end? J. Cell Sci. 122, 3605–3612.
[5] Grudnik, P., Bange, G. and Sinning, I. (2009) Protein targeting by the signal recognition particle. Biol. Chem. 390, 775–782.
[6] Cross, B.C., Sinning, I., Luirink, J. and High, S. (2009) Delivering proteins for export from the cytosol. Nat. Rev. Mol. Cell Biol. 10, 255–264.
[7] Kutay, U., Hartmann, E. and Rapoport, T.A. (1993) A class of membrane proteins with a C-terminal anchor. Trends Cell Biol. 3, 72–75.
[8] Stefanovic, S. and Hegde, R.S. (2007) Identification of a targeting factor for posttranslational membrane protein insertion into the ER. Cell 128, 1147–1159.
[9] Favaloro, V., Spasic, M., Schwappach, B. and Dobberstein, B. (2008) Distinct targeting pathways for the membrane insertion of tail-anchored (TA) proteins. J. Cell Sci. 121, 1832–1840.
[10] Mateja, A., Szlachcic, A., Downing, M.E., Dobosz, M., Mariappan, M., Hegde, R.S. and Keenan, R.J. (2009) The structural basis of tail-anchored membrane protein recognition by Get3. Nature 461, 361–366.
[11] Bozkurt, G. et al. (2009) Structural insights into tail-anchored protein binding and membrane insertion by Get3. Proc. Natl. Acad. Sci. USA 106, 21131–21136.
[12] Suloway, C.J., Chartron, J.W., Zaslaver, M. and Clemons Jr., W.M. (2009) Model for eukaryotic tail-anchored protein binding based on the structure of Get3. Proc. Natl. Acad. Sci. U.S.A. 106, 14849–14854.
[13] Hu, J., Li, J., Qian, X., Denic, V. and Sha, B. (2009) The crystal structures of yeast Get3 suggest a mechanism for tail-anchored protein membrane insertion. PLoS One 4, e8061.
[14] Yamagata, A., Mimura, H., Sato, Y., Yamashita, M., Yoshikawa, A. and Fukai, S. (2010) Structural insight into the membrane insertion of tail-anchored proteins by Get3. Genes Cells 15, 29–41.
[15] Schuldiner, M., Metz, J., Schmid, V., Denic, V., Rakwalska, M., Schmitt, H.D., Schwappach, B. and Weissman, J.S. (2008) The GET complex mediates insertion of tail-anchored proteins into the ER membrane. Cell 134, 634–645.
[16] Jonikas, M.C. et al. (2009) Comprehensive characterization of genes required for protein folding in the endoplasmic reticulum. Science 323, 1693–1697.
[17] Fleischer, T.C., Weaver, C.M., McAfee, K.J., Jennings, J.L. and Link, A.J. (2006) Systematic identification and functional screens of uncharacterized proteins associated with eukaryotic ribosomal complexes. Genes Dev. 20, 1294–1307.
[18] Kobe, B. and Kajava, A.V. (2000) When protein folding is simplified to protein coiling: the continuum of solenoid protein structures. Trends Biochem. Sci. 25, 509–515.
[19] D'Andrea, L.D. and Regan, L. (2003) TPR proteins: the versatile helix. Trends Biochem. Sci. 28, 655–662.
[20] Groves, M.R. and Barford, D. (1999) Topological characteristics of helical repeat proteins. Curr. Opin. Struct. Biol. 9, 383–389.
[21] Hierro, A. et al. (2007) Functional architecture of the retromer cargo-recognition complex. Nature 449, 1063–1067.
[22] Holm, L., Kaariainen, S., Rosenstrom, P. and Schenkel, A. (2008) Searching protein structure databases with DaliLite v.3. Bioinformatics 24, 2780–2781.
[23] Collins, B.M., McCoy, A.J., Kent, H.M., Evans, P.R. and Owen, D.J. (2002) Molecular architecture and functional model of the endocytic AP2 complex. Cell 109, 523–535.
[24] Goldenberg, S.J., Cascio, T.C., Shumway, S.D., Garbutt, K.C., Liu, J., Xiong, Y. and Zheng, N. (2004) Structure of the Cand1–Cul1–Roc1 complex reveals regulatory mechanisms for the assembly of the multisubunit cullin-dependent ubiquitin ligases. Cell 119, 517–528.
[25] Rice, L.M. and Brunger, A.T. (1999) Crystal structure of the vesicular transport protein Sec17: implications for SNAP function in SNARE complex disassembly. Mol. Cell 4, 85–95.
[26] Groves, M.R., Hanlon, N., Turowski, P., Hemmings, B.A. and Barford, D. (1999) The structure of the protein phosphatase 2A PR65/A subunit reveals the conformation of its 15 tandemly repeated HEAT motifs. Cell 96, 99–110.
[27] Krogan, N.J. et al. (2006) Global landscape of protein complexes in the yeast *Saccharomyces cerevisiae*. Nature 440, 637–643.
[28] Abe, Y., Shodai, T., Muto, T., Mihara, K., Torii, H., Nishikawa, S., Endo, T. and Kohda, D. (2000) Structural basis of presequence recognition by the mitochondrial protein import receptor Tom20. Cell 100, 551–560.
[29] Gatto Jr., G.J., Geisbrecht, B.V., Gould, S.J. and Berg, J.M. (2000) Peroxisomal targeting signal-1 recognition by the TPR domains of human PEX5. Nat. Struct. Biol. 7, 1091–1095.
[30] Scheufler, C., Brinker, A., Bourenkov, G., Pegoraro, S., Moroder, L., Bartunik, H., Hartl, F.U. and Moarefi, I. (2000) Structure of TPR domain-peptide complexes: critical elements in the assembly of the Hsp70–Hsp90 multichaperone machine. Cell 101, 199–210.

[31] Zhang, Y. and Chan, D.C. (2007) Structural basis for recruitment of mitochondrial fission complexes by Fis1. Proc. Natl. Acad. Sci. USA 104, 18526–18530.
[32] Chang, Y.W., Chuang, Y.C., Ho, Y.C., Cheng, M.Y., Sun, Y.J., Hsiao, C.D. and Wang, C. (2010) Crystal structure of Get4/Get5 complex and its interactions with Sgt2, Get3 and Ydj1. J. Biol. Chem. (epub Jan 27).
[33] Van den Berg, B., Clemons Jr., W.M., Collinson, I., Modis, Y., Hartmann, E., Harrison, S.C. and Rapoport, T.A. (2004) X-ray structure of a protein-conducting channel. Nature 427, 36–44.
[34] Costanzo, M. et al. (2010) The genetic landscape of a cell. Science 327, 425–431.